

## ABSTRACT

A method for determining the concentration of a substrate in a sample solution using an electrode system comprising a working electrode and a counter electrode, both being formed on an electrically insulating base plate, and a reaction layer which contains at least an oxidoreductase and an electron mediator and is formed on the electrode system to electrochemically measure a reduced amount of the electron mediator resulting from enzyme reaction in the reaction layer, wherein a third electrode is formed as an interfering substance detecting electrode somewhere apart from the reaction layer to detect supply of the sample solution on the basis of an electrical change between the counter electrode and the third electrode. A current flowing between the counter electrode and the third electrode is measured which is taken as a positive error. Subsequently, voltage application between the counter electrode and the third electrode is released and a voltage for oxidizing the reduced form electron mediator is applied between the working electrode and the counter electrode to measure a current flowing between the two electrodes. Influences of any interfering substance such as easy-to-oxidize substance are reduced, whereby a highly reliable value of substrate determination can be obtained.